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# Studies on the Propionibacterium. (IV)

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From the above results it seems probable that the decrease of the coefficient of friction is the chief factor for the decrease of the torsional resistance of the viscose rayon yarn treated by this oiling agents, considering the fact that the Young's modulus is not decreased by those treatments.

In the case of raw silk, it was found, as reported in the previous work, that the torsional resistance decreased with oiling. Another experiment was carried out using polyamide "Amilan" monofilament and the decrease of both the torsional resistance and the coefficient of friction was recognized by using Pansofter as an oiling agent. In the case of Amilan, it seems that not only the coefficient of friction is decreased but also the softening of the fiber itself is resulted by oiling.

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## 24. Studies on the *Propionibacterium*. (IV)

Hideo KATAGIRI and Yoshio ICHIKAWA

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Delwhiche (*J. Bact.* 56, 811 (1948)) has shown that *Propionibacterium pentosaceum* converts succinic acid into propionic acid and carbon dioxide at pH 5.2. Fromageot and Bost (*Enzymologia*, 4, 225 (1938)) obtained the same result in presence of glucose at pH 6.4.

It is found in the present paper that yeast extract is necessary for decarboxylation of succinate by *P. arabinosum*, and that the effect of yeast extract appears in its inorganic constituents.

The medium used for growing bulk contained 1 g. of glucose, 1 g. of peptone, and 0.5 g. of Difco Yeast Extract in 200 ml.; the initial pH was 7.0. The medium was inoculated with 10 ml. of 24 hrs. subculture of *P. arabinosum*. After 36 hrs. incubation at 30°, the bacterial cells were centrifuged; washed with 100 and then 50 ml. phosphate buffer and finally suspended in 5 ml. of water. The yield of the fresh cells were found to be between 0.8-0.9 g. (nearly 86 mg. on dry basis). The fresh suspension thus obtained was used in every experiments.

The decarboxylase activity was determined by manometric measurement of carbon dioxide evolved. As a rule, cell suspension (0.5 ml.), phosphate buffer (final concentration was 1/30 *M*) and any other additions except substrates were placed in the main chamber of the cup, while the substrate solution was put into the side chamber. The total volume of fluid in the manometer cup was adjusted to be 3.0 ml.; air space in the cup was displaced by CO<sub>2</sub>, and the temperature of water bath was kept at 30°.

Succinate is not decarboxylated at pH 6.4 even when yeast extract and glucose are added.

At pH 5.2, decarboxylation of succinate was very much accelerated by addition of glucose and yeast extract, while no remarkable acceleration was observed by the addition of any of them, since gas evolution slightly increased by yeast extract and any effect was never pointed out by glucose as is shown in Table 1.

Table 1.

Substrates added	CO <sub>2</sub> (μl.) evolved in		
	10	20	30 min.
No substrate	0	7	13
Na-succinate 10 <sup>-2</sup> <i>M</i>	0	7	14
Succinate 10 <sup>-2</sup> <i>M</i> + glucose 10 <sup>-3</sup> <i>M</i>	33	52	53
Glucose 10 <sup>-3</sup> <i>M</i>	31	51	54
Succinate 10 <sup>-2</sup> <i>M</i> + yeast extract 0.3ml.	36	54	57
Yeast extract 0.3ml.	26	36	50
Succinate 10 <sup>-2</sup> <i>M</i> + glucose 10 <sup>-3</sup> <i>M</i> + yeast extract 0.3ml.	60	86	103
Glucose 10 <sup>-3</sup> <i>M</i> + yeast extract 0.3ml.	49	63	75

No effect on the decarboxylation of succinate was again observed even when the cells were pre-incubated with glucose for 30 min. prior to the addition of succinate. It will be seen in Table II that the effect of yeast extract was revealed by yeast ash extract (ashes from 2g. dried yeast was dissolved in 10 ml. of phosphate buffer of pH 5.2 for 24 hrs. at room temperature) and by Mg<sup>++</sup> or Mn<sup>++</sup>.

Table 2. Succinate 10<sup>-2</sup>*M*, glucose 10<sup>-3</sup>*M*.

Further additions	CO <sub>2</sub> (μl.) evolved in	
	15	30 min.
Yeast extract 0.3ml.	42	52
Yeast ash extract 0.3ml.	64	77
MnSO <sub>4</sub> 1/100 <i>M</i>	50	61
MnCl <sub>2</sub> 1/150 <i>M</i>	49	64
MgCl <sub>2</sub> 1/150 <i>M</i>	31	49

The effect of phosphate and the optimum concentration of inorganic matters were investigated by using 1/30 *M* acetate buffer, and found that phosphate altered the initial velocity of the decarboxylation of succinate, and that the decarboxylation was accelerated by higher concentrations of Mn<sup>++</sup> or Mg<sup>++</sup>. (See Table 3).

Table 3. Succinate  $10^{-2}M$ : glucose  $10^{-3}M$ .

Further additions	CO <sub>2</sub> ( $\mu$ l.) evolved in	
	10	30 min.
MnCl <sub>2</sub> $10^{-2}M$	60	81
MnCl <sub>2</sub> $10^{-3}M$	51	71
MnCl <sub>2</sub> $10^{-4}M$	21	33
MgCl <sub>2</sub> $10^{-2}M$	41	59
MgCl <sub>2</sub> $10^{-3}M$	30	41

## 25. Polarographic Studies on the Urinal Colloids. (II)

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In the preceding paper (T. Sasai, M. Egawa, Y. Kumahara and H. Hayami: *This Bulletin*, 29, 15, 1952) it was reported that the urinal components showing the protein double wave were precipitated by alcohol-addition and were identical with the mucoprotein; furthermore it was indicated that this mucoprotein was related to the occurrence of the so-called lability test in serum or in urine such as Weltmann test or Donaggio test. The detail of the latter fact will be stated in the other paper. In this report the experimental results are shown which indicated the difference between two samples of mucoprotein, i. e. the one obtained from normal urine, the other from the patient with liver cancer.

1. Comparison of two curves which express the relationship between the concentration of mucoprotein and their wave-height: Both samples showed similar curves agreeing with the one of isothermal adsorption. Approximately up to the conc. of 0.01% they are almost linear and coincident each other. In the region over 0.01% they are separated, in the manner that the wave from healthy mucoprotein is higher than that of liver cancer, for example in 0.02% the former corresponds to 168mm., the latter to 132mm. Beyond the conc. of 0.07% they reached the constant value, that is to say the limiting value of the adsorption curve: 270mm. and 145mm., respectively.

2. When the hydrolysed samples were examined to measure the height as the cystin waves using Brdička's method (Brdička, R.: *Collection Czech.* 5, 233, (1933)), difference was found between them: 0.04% of normal sample gave 58mm., while the corresponding value of cancer-sample 54mm., if taken G.S. as 1/100. Similar differences were also resulted in other three samples obtained from the patients of gastric cancer, pulmonary tuberculosis and portal cirrhosis.